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CHILINA IGUAZUENSIS (GASTROPODA: CHILINIDAE), NEW SPECIES FROM IGUAZÚ NATIONAL PARK, ARGENTINA

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ABSTRACT

The Chilinidae comprises 16 species currently cited for Argentina, mostly distributed in Patagonia. All original descriptions of these species have been based on shell characters, and their internal anatomy is poorly known. Here a new species, ***Chilina iguazuensis***, is described, including shell, radula, and reproductive and nervous systems. This species, found in Iguazú National Park, Misiones Province, Argentina, in the Upper Iguazú River rapids, has the following distinctive characteristics: aperture length equal to last whorl length; central radular tooth asymmetric and bicuspid, with both cusps serrated; and prepuce length 60% of penis sheath length. ***Chilina iguazuensis*** is endemic in Iguazú National Park, along with *Chilina megastoma,* which inhabits waterfalls in the same river.

Key words: ***Chilina iguazuensis***, n. sp., Chilinidae, anatomy, Argentina.

INTRODUCTION

The family Chilinidae Dall, 1870 (Gastropoda, Pulmonata, Basommatophora), is exclusive to South America, occurring from the Tropic of Capricorn to Cape Horn and the Falkland Islands. The family comprises a single genus, *Chilina* Gray, 1828 (type species *Auricula* (*Chilina*) *fluctuosa* Gray, 1828, Chile), with about 32 species, 16 of which have been recorded in Argentina (Castellanos & Miquel, 1980, 1991; Castellanos & Gaillard, 1981; Castellanos & Landoni, 1995). Original species descriptions are restricted to shell characters (Hidalgo, 1880; Marshall, 1924, 1933; Hylton Scott, 1958, among others), and very few subsequent works have dealt with internal anatomy (Haeckel, 1911; Duncan, 1960a, b, 1975; Harry, 1964; Brace, 1983; Ituarte, 1997). Miquel (1984, 1987) analyzed the penial complex of eight species from Argentina, Brazil, Chile, and Uruguay, and concluded that no microanatomical differences could be found at this level. Based on characteristics of Chilean species, Valdovinos & Stuardo (1995) rearranged the family systematics and proposed that the nervous system had the only reliable characters enabling definitions of supraspecific taxa.

From an evolutionary perspective, the family Chilinidae is among the most primitive pulmonate gasteropods, and one of the first that conquered the aquatic environment. This is reflected in such primitive characters as a streptoneurous nervous system, horizontal lamellar tentacles, non-contractile pneumostome, and incomplete division of male and female ducts (Hubendick, 1947, 1978; Duncan, 1960a; Harry, 1964; Brace, 1983). More recently, based on analyses of 28S rRNA sequences, Dayrat et al. (2001) confirmed the monophyly of Hygrophyla, including the Chilinidae at the base of this clade.

The genus *Chilina* has the following distinctive characteristics: Shell: oval, oblong to ventricose, with last whorl expanded, spire erect or immersed, whorls generally convex (sometimes carinate or angulose), aperture oval with columellar and parietal margins callous, with up to two oblique, tooth-like folds. Periostracum always present, olive-yellow, with dark brown longitudinal zigzag bands. Digestive system: Salivary glands slightly lobed; esophagus rather narrow; muscular stomach forming a caecum; anus opening to large mantle lobe; radula with numerous V-shaped tooth rows, with one central tooth and numerous lateral and marginal teeth laterally and posteriorly to central tooth. Reproductive system: genital system that is primitive among the Basommatophora because of the incomplete separation

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TABLE 1. Number of specimens collected during different months at each rapid where ***Chilina iguazuensis*** was recorded, at Iguazú National Park, Argentina.

	Ñandú 25°42'S, 54°25'W	Tacuara 25°36'S, 54°21'W	Mbigüa 25°41'S, 54°26'W	León 25°36'S, 54°14'W	Apepú 25°33'S, 54°17'W	Irene 25°38'S, 54°23'W
MLP	12526-28	12529-30	12532	12534	12533	12531
Feb. 2004	70					
June 2004		20				
Sept. 2004	25		2			
Feb. 2005		17		10	15	16
June 2005	20					
Dec. 2005	65		16			

of male and female reproductive ducts, absence of fertilization chamber, and presence of very numerous penial spines; accessory seminal receptacle and calcareous granules in the vaginal lumen (Haeckel, 1911; Duncan 1960a, b; Harry 1964; Castellanos & Gaillard, 1981; Miquel, 1984).

Here we describe a new species of this family, ***Chilina iguazuensis***, from Iguazú National Park, Argentina; we provide descriptions of the shell and soft body parts, in particular the radula and internal organs.

MATERIALS AND METHODS

Specimens were collected during 2004 and 2005 at Iguazú National Park, Misiones Province, Argentina (Fig. 1, Table 1). Soft parts were separated from the shell for subsequent processing, after relaxation in 10% Nembutal solution for 12 h, and fixed in modified Raillet-Henry solution for freshwater animals – 93% distilled water, 2% glacial acetic acid, 5% formaldehyde, and 6 g sodium chloride per liter. Radulae were separated from the buccal mass and cleaned with sodium hypochlorite (Clorox).

Six shell measurements were taken (Fig. 2): total length (TL), last whorl length (LWL), aperture length (AL), total width (TW), aperture width (AW), aperture projection (AP) and number of teeth, following Gutiérrez et al. (1994) and Martín (2003). Radulae (n = 6) were observed under a scanning electron microscope at Museo de La Plata, Facultad de Ciencias Naturales y Museo, Universidad Nacional de

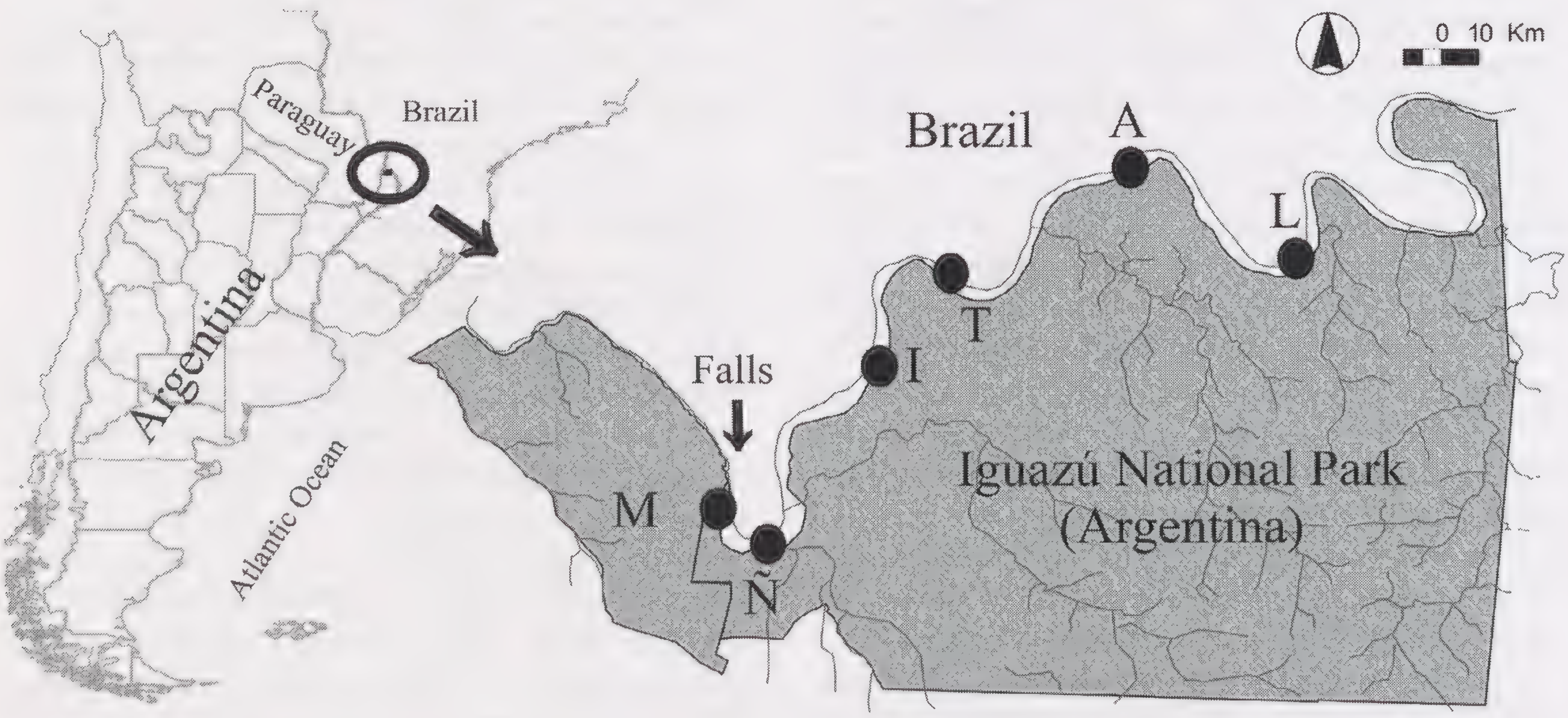

FIG. 1. Rapids where ***Chilina iguazuensis*** was recorded in Iguazú River, within Iguazú National Park, Argentina. A: Apepú; I: Irene; L: León; M: Mbigüa; Ñ: Ñandú; T: Tacuara.

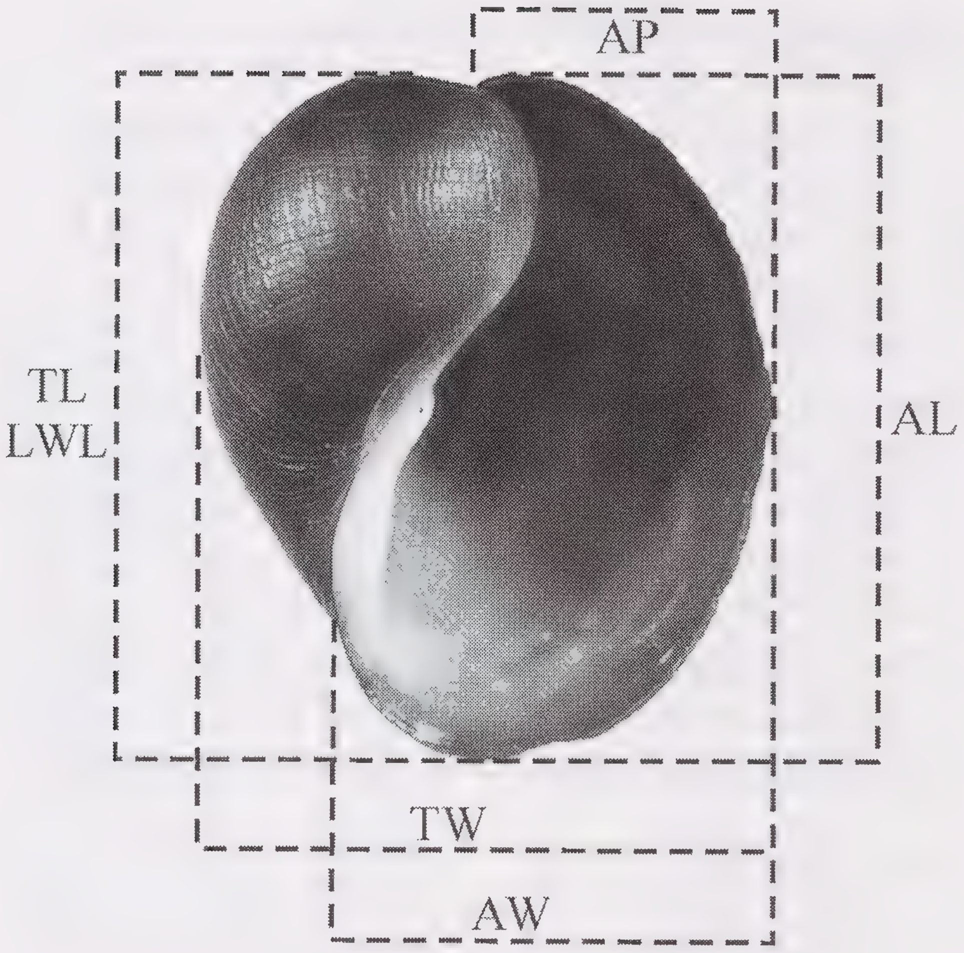

FIG. 2. Shell measurements used for ***Chilina iguazuensis***: TL: total length; LWL: last whorl length; AL: aperture length; TW: total width; AW: aperture width; AP: aperture projection.

La Plata. Internal anatomy (n = 11) was analyzed using a stereoscopic binocular microscope Leica MZ6 with camera lucida.

Shell characters were compared to the original description of *Chilina megastoma* Hylton Scott, 1958, a species that also occurs in Iguazú National Park, but inhabits waterfalls of the Iguazú River basin. Anatomical traits were compared with the work of Ituarte (1997) about the latter species, as well as with our own measurements from 11 *C. megastoma* specimens collected at Arrechea Fall, Iguazú National Park, prepared following the above methodology.

Average measurements of soft parts were divided by the length of the last whorl, in order to obtain size-free variables that facilitate comparisons among different individuals and species. Length of last whorl was preferred over dimensions of soft parts because it was the most consistent measurement among individuals, and it was not affected by relaxation techniques.

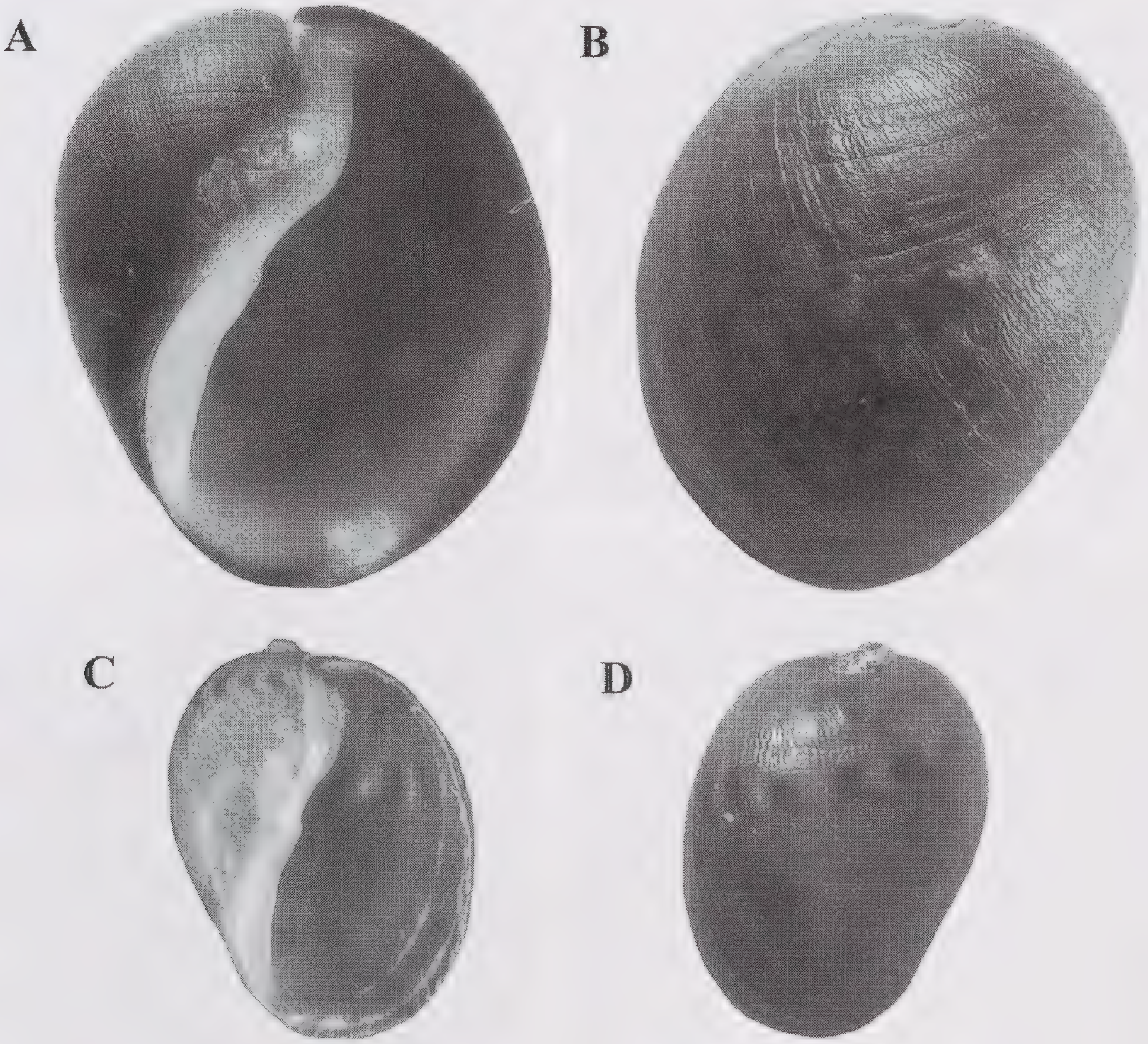

FIG. 3. Shell of ***Chilina iguazuensis***, n. sp. A, B: Adult specimen (Holotype) (18.08 mm); C, D: Juvenile specimen (6.27 mm). A, C = apertural view; B, D = dorsal view.

RESULTS

Chilina iguazuensis, sp. nov.

(Figs. 1–7)

Type Locality

Upper Iguazú River, Iguazú National Park, Misiones Province, Argentina.

Type Material

Holotype (MLP: 12526) and paratypes (MLP: 12527, 12528) at Museo de La Plata; paratypes (MACN-In 37175) at Museo Argentino de Ciencias Naturales "Bernardino Rivadavia".

Etymology

The specific name refers to the type locality, Iguazú River in Iguazú National Park.

Diagnosis

Shell globular, spire immersed in adult specimens (visible only in specimens less than 7 mm long), length of aperture equal to length of last whorl; interior of shell markedly iridescent. Radula: 57–65 teeth rows; 51–63 teeth per half-row; central tooth asymmetrical, bicuspid, both cusps serrated and second lateral tooth tetracuspid. Reproductive system: prepuce length 60% of penis sheath length; secondary bursa copulatrix tubular, short. Nervous system: mean distance between left pleural and parietal ganglia 1.39 mm (SD: 0.97), approximately 7% of last whorl length.

Description

Shell (Fig. 3): Shell strong, oval, globular. Color purplish. Spire immersed, except in very small specimens (< 7mm). Last whorl very large, dilated, length equal to aperture

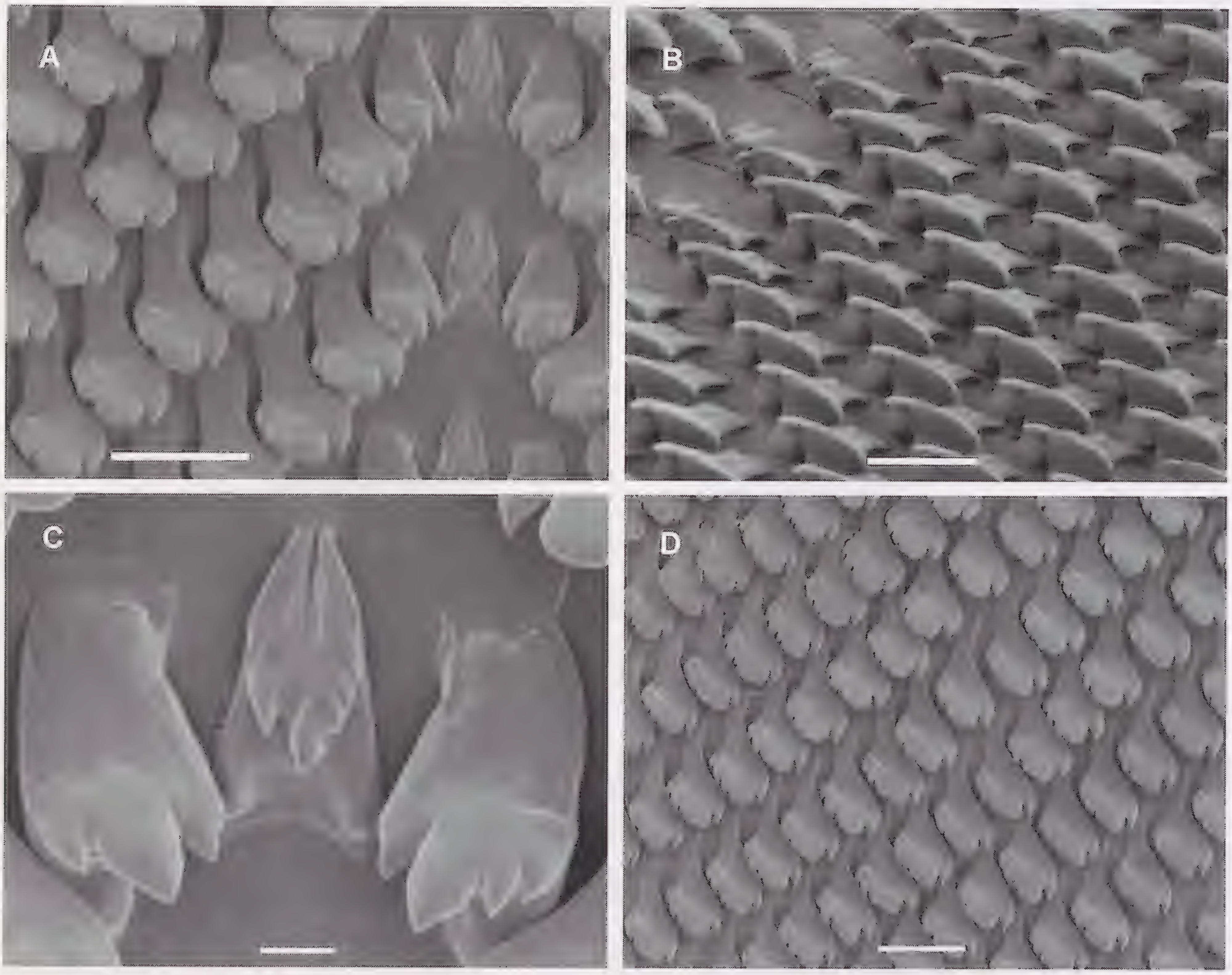

FIG. 4. Radula of ***Chilina iguazuensis***. A: General view; B: Lateral view; C: Central tooth and first lateral teeth; D: marginal teeth. Scale bars = 10 µm.

TABLE 2. Shell measurements of ***Chilina iguazuensis***. All measurements in mm. Sp: specimen; TL: total length; LWL: last whorl length; AL: aperture length; TW: total width; AW: aperture width; AP: aperture projection.

	Date	Sp.	TL	LWL	AL	TW	AW	AP
Ñandú								
Holotype MLP12526	Feb. 2004	3	18.08	18.08	18.08	14.87	12.45	7.14
Paratype MLP12527	Feb. 2004	1	21.81	21.81	21.21	17.90	13.46	6.20
Paratype MLP12527	Feb. 2004	2	16.62	16.62	16.62	13.60	11.00	7.42
Paratype MLP12527	Feb. 2004	4	18.39	18.39	18.39	15.22	11.78	6.96
Paratype MLP12527	Feb. 2004	5	15.05	15.05	15.05	12.46	10.25	6.06
Paratype MLP12527	Feb. 2004	6	16.35	16.35	16.35	13.37	10.10	6.91
Paratype MLP12528	Sept. 2004	1	20.24	20.24	20.24	16.70	11.65	6.48
Paratype MLP12528	Sept. 2004	2	20.66	20.66	20.66	18.82	13.15	8.99
Paratype MLP12528	Sept. 2004	3	22.16	22.16	22.16	18.37	13.64	9.08
Paratype MLP12528	Sept. 2004	4	19.76	19.76	19.76	17.84	12.36	8.07
Paratype MLP12528	Sept. 2004	5	17.71	17.71	17.71	15.23	11.89	7.27
Paratype MLP12528	Sept. 2004	6	17.63	17.63	17.63	14.46	11.15	6.94
Paratype MLP12528	Sept. 2004	7	21.01	21.01	21.01	17.14	12.51	7.50
Tacuara								
MLP 12529	June 2004	1	20.56	20.56	19.66	17.15	12.96	5.88
MLP 12529	June 2004	2	12.13	12.13	12.24	9.96	8.09	5.13
MLP 12529	June 2004	3	12.22	12.22	12.22	9.91	7.37	4.00
MLP 12529	June 2004	4	23.91	23.91	23.10	21.22	14.10	7.50
MLP 12529	June 2004	5	22.95	22.95	22.14	17.75	13.12	7.13
MLP 12529	June 2004	6	24.12	24.12	22.10	19.49	12.50	6.83

length. Well-defined growth lines crossed by spiral grooves, forming reticulate shell ornamentation. A zigzag colored band on last whorl of juveniles, absent in adults. Aperture large, peristome outline oval. Labrum simple. Columellar callosity white, straight. One columellar tooth. Interior markedly iridescent. Parietal callosity weak, with soft white coloration. Two whorls. Maximum size recorded 24.12 mm total length. Table 2 lists shell measurements for 19 specimens.

Digestive System: General morphology follows the basic plan described above for the genus. In this section we provide a description of radular morphology.

Radula with numerous tooth rows arranged in V-shape, as in all species of the family (Fig. 4A). All teeth except central united to membrane by base and an anterior projection, base directed forward of radula and projection backward, and both situated at base of cusps (Fig. 4B). Six individuals ranging between 12.13 and 22.95 mm total length examined. Mean estimated number of rows 58; number of rows 57 (12.13 mm specimen) to 65 (19.2 mm). Number of teeth per half-row based on counts of first rows (4–9). Mean number of teeth per half-row (not including central tooth) 51, ranging from 43 (12.13 mm specimen) to 63 (22.95 mm specimen). Central tooth (Fig. 4C) asymmetrical, bicuspid, with one cusp slightly displaced to right; this latter cusp bifurcated in some specimens. External margins of both cusps serrate. Free part of central tooth not flat but with anteroposterior groove originating from inward folding of basal plate, becoming narrower and deeper posteriorly (Fig. 4C). A protuberance at each margin of anterior area of tooth base.

First lateral tooth tricuspid, with larger dagger-shaped mesocone higher and broader than other cusps (Fig. 4C). Ectocone of left tooth bifurcated in one individual. First lateral tooth slightly curved toward central tooth. Second lateral tooth tetracuspid, with smaller, divided ectocone (fourth cusp) and undivided endocone and mesocone. Contiguous teeth with more rounded appearance due to smaller cusps. Last marginal teeth simple, with poorly developed cusps, and ectocone divided into three minor cusps (Fig. 4D).

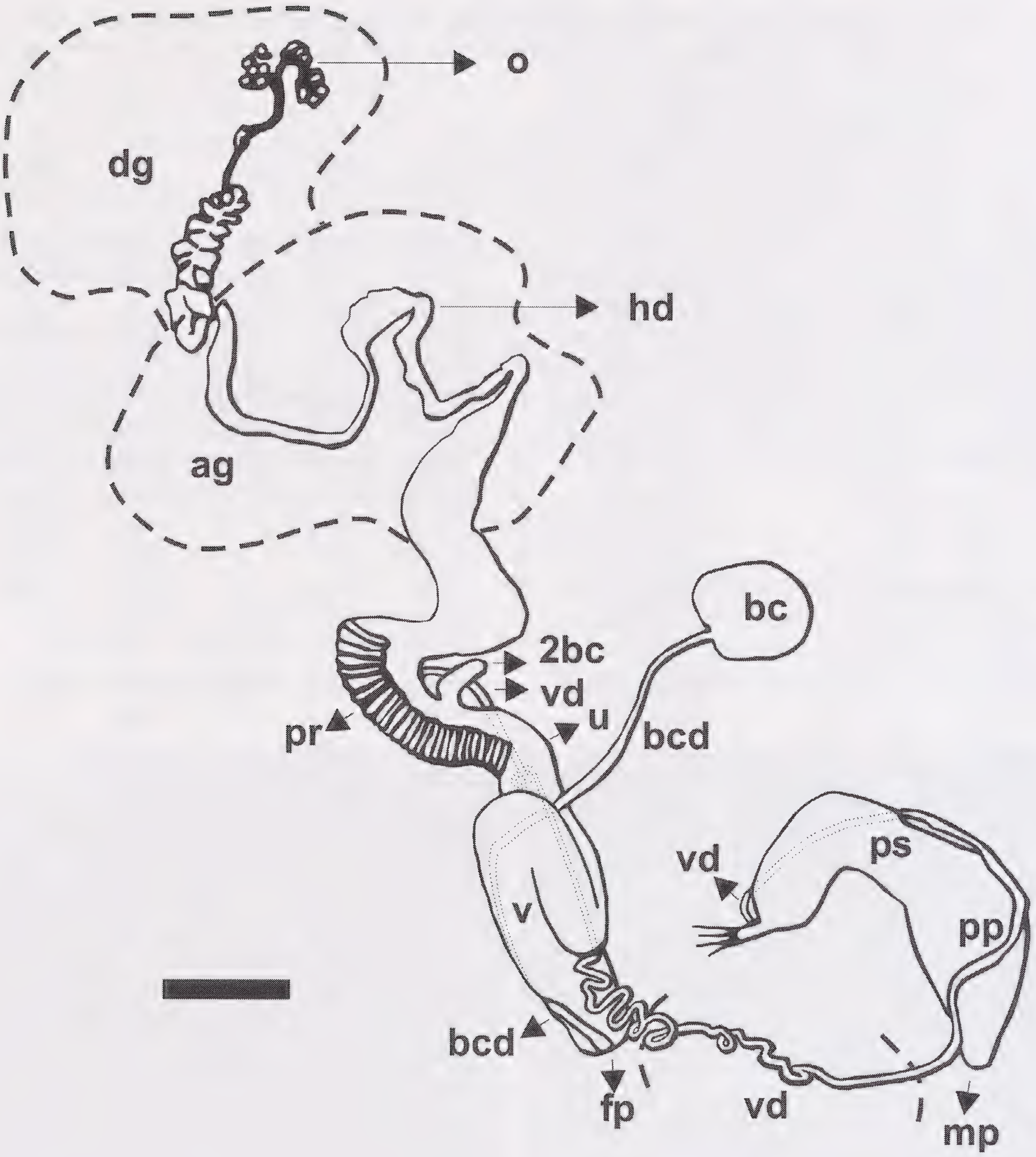

FIG. 5. Diagram of reproductive system of ***Chilina iguazuensis***: ag: albumen gland; vd: vas deferens; dg: digestive gland; bc: bursa copulatrix; bcd: bursa copulatrix duct; fp: female pore; hd: hermaphrodite duct; mp: male pore; o: ovotestis; pp: prepuce; pr: prostate; ps: penis sheath; u: uterus; v: vagina; 2bc: secondary bursa copulatrix. Dotted line: body wall. Scale bar = 2 cm.

Radular Formula: [51/(3-5) + 1/2] 57-65 [= number of right and left teeth/(number of cusps) + number of central teeth/number of cusps] extreme numbers for transversal rows.

Reproductive System (Fig. 5): Ovotestis (hermaphroditic gonad) and a common duct anterior to the separation of both systems. Female and male reproductive elements are described separately. Distinctive characteristics of ***C. iguazuensis***:

Female Genital System: Bursa copulatrix duct emerging from anterodorsal vagina and extending alongside intestine, then passing under vagina and over uterus-vagina complex toward visceral mass; finally opening into flat oval bursa copulatrix next to ventricle. Secondary bursa copulatrix arising at base of uterus (oviduct); short (21% of length of the bursa copulatrix duct) and cylindrical, not expanded distally.

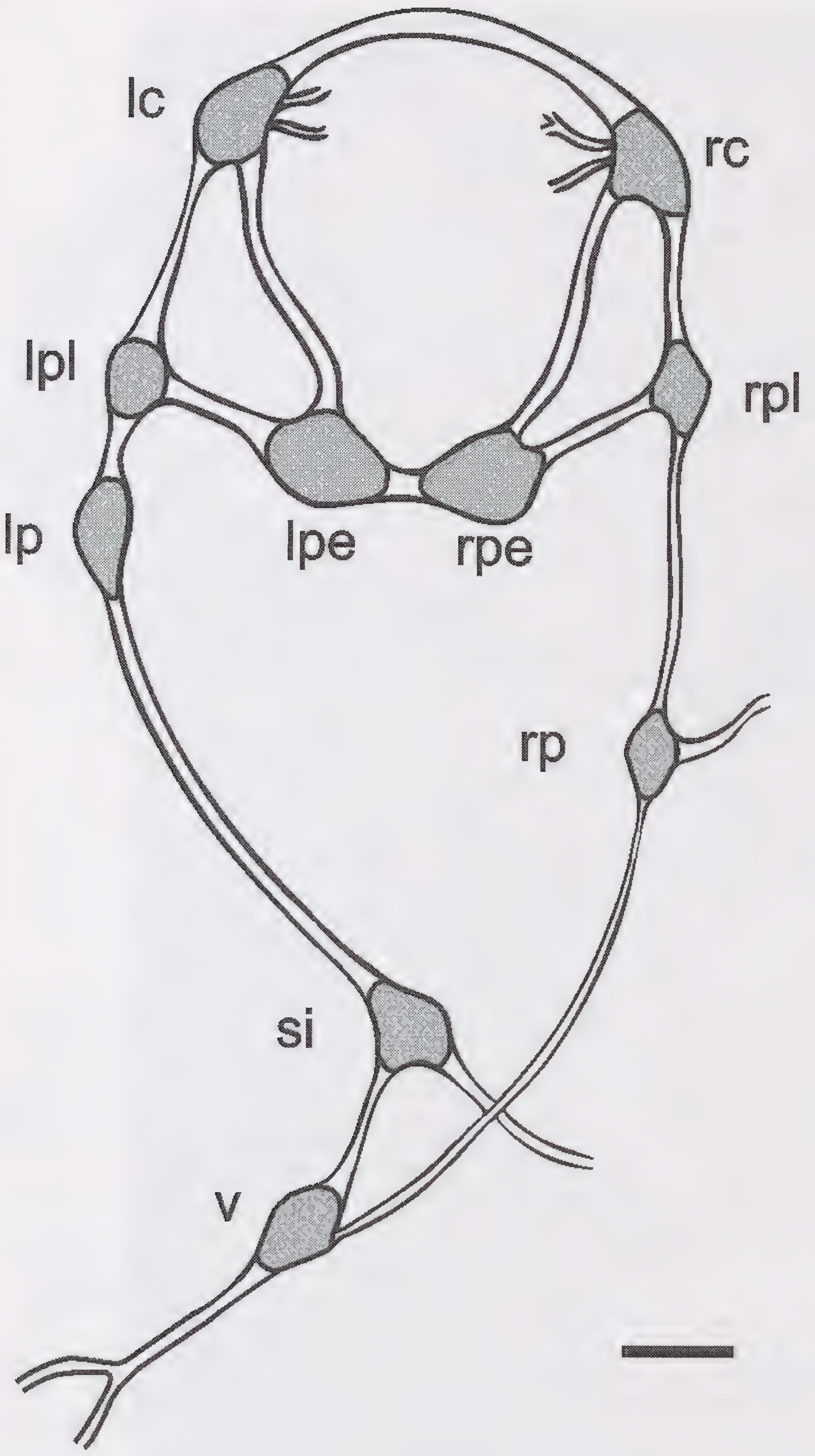

FIG. 6. Diagram of nervous system of ***Chilina iguazuensis***. Abbreviations for ganglia: lc: left cerebral; lpe: left pedal; lp: left parietal; lpl: left pleural; rc: right cerebral; rpe: right pedal; rp: right parietal; rpl: right pleural; si: subintestinal; v: visceral. Scale bar = 1 mm.

Male Genital System: Vas deferens emerging from prostate; prostrate surrounding albumen gland (obvious in recently fixed specimens due to yellowish coloration). Vas passing under uterus and vagina, twisting upon itself, and finally located over vagina, once past final bend of the latter (Fig. 5). Vas deferens running laterally to buccal mass and not above vagina, coiling upon itself at this sector and making four loops; then entering body wall on right side with several minor turns and twists; this section twice as long as distance between vaginal and male pores. Vas deferens emerging from body wall at penial complex level and extending toward latter, crossing above prepuce. At this section vas extending alongside penis sheath in straight trajectory, and entering the latter near buccal mass. Prepuce length 60% of penis sheath length. Structure of prepuce, penis and penis sheath similar to those described by Ituarte (1997) for *Chilina megastoma*.

Nervous System (Figs. 6, 7; Table 3): Pedal and cerebral ganglia connected by commissures and connectives, forming anterior nerve ring located at anterior half of buccal mass, only slightly posterior to beginning of esophagus. Long connective (ratio: 13.53 of length of the last whorl) linking right pleural ganglion to right parietal ganglion. Right parietal ganglion giving rise to two nerves, one to osphradium and one long, very thin connective to visceral ganglion behind posterior nerve ring. Two connectives on left side of anterior nerve ring, longer than those on right side, connecting left cerebral and pedal ganglia to pleural ganglion. Small connective (ratio: 4.79 of last whorl length) from pleural ganglion to left of parietal ganglion; this connective 65% shorter than right-side counterpart. Long connective (ratio: 18.43 of last whorl length) linking left parietal ganglion to subintestinal ganglion, located above posterior half of columellar muscle. One very short connective (ratio: 6.28 of last whorl length) linking subintestinal ganglion to visceral ganglion and closing posterior nerve ring. Two nerves arising from visceral ganglion toward visceral mass. One large nerve extending to right from subintestinal ganglion through columellar muscle to innervate distal-most vagina and accesory pneumostome. Pleurovisceral connectives with incomplete torsion characteristic of the genus. Table 3 shows proportions between length of connectives and length of last whorl.

Distribution

The new species was detected only at the type locality, Iguazú National Park, in the rapids of Upper Iguazú River listed in Table 1.

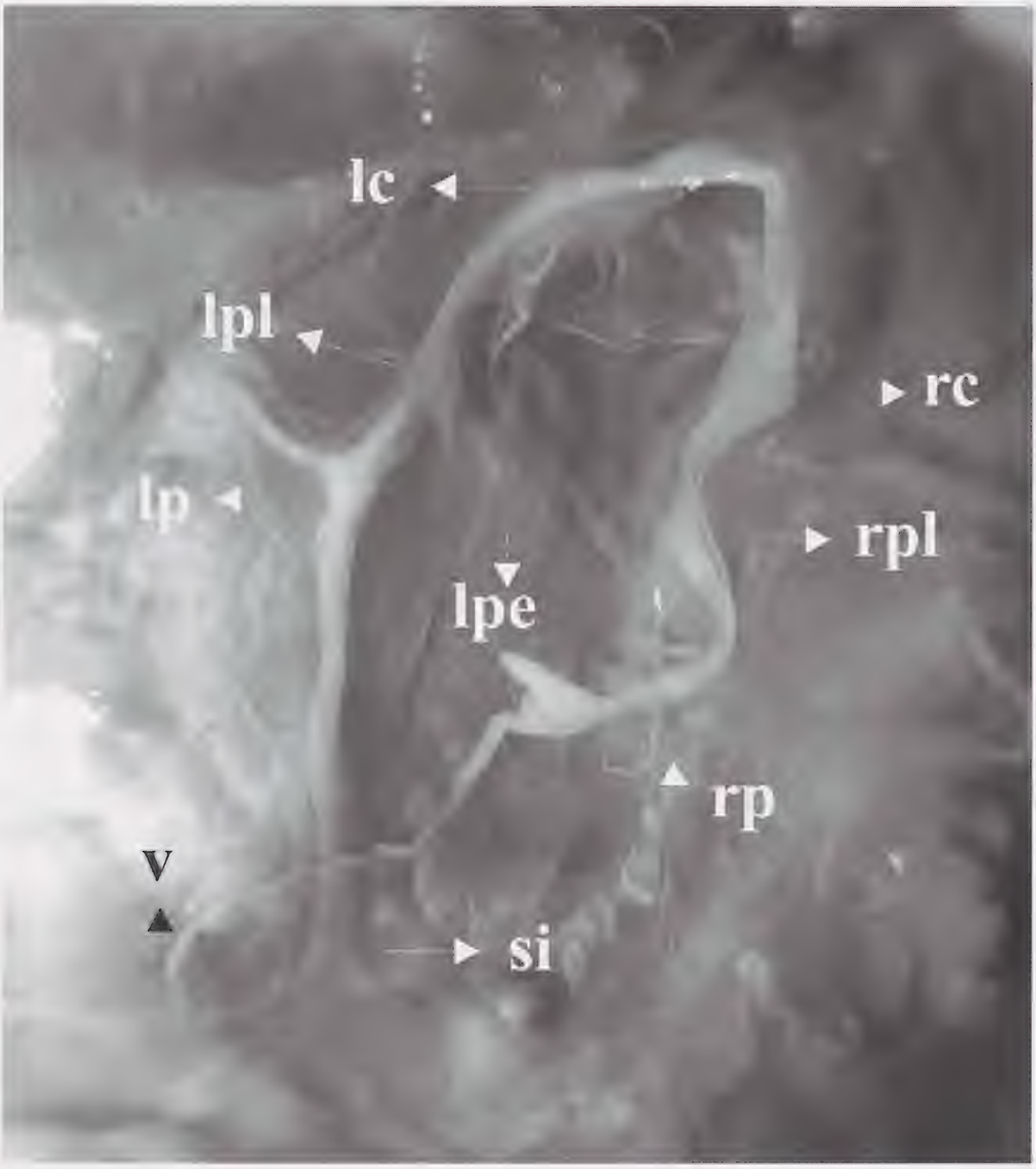

FIG. 7. Nervous system of ***Chilina iguazuensis*** in situ.

DISCUSSION

The species closest to ***C. iguazuensis*** based on shell, anatomical and distributional characteristics is *Chilina megastoma* (Fig. 8). The populations of ***C. iguazuensis*** were recorded along the course of the upper Iguazú River, mainly in rapids. In contrast, *C. megastoma* only occurs in wet zones formed by the spray of the Iguazú River in the vicinity of Iguazú National Park, Argentina-Brazil.

So far, two other *Chilina* species are known for areas geographically close to the range of the new species: *Chilina gallardoi* Castellanos & Gaillard, 1981, from Uruguay River, which is not connected to the Iguazú River; and *Chilina guaraniana* Castellanos & Miquel, 1980, from the Paraná River. The latter species was described based on materials collected in 1935 and not recorded since. The populations of both these species are located about 300 km south of Iguazú National Park. The internal anatomy of these two species is as yet unknown. Both are markedly different from ***C. iguazuensis*** and *C. megastoma* in shell morphology, with the aperture not greatly developed in either *C. gallardoi* or *C. guaraniana*. In addition, the aperture of *C. gallardoi* has two well-developed (columellar and parietal) teeth, and *C. guaraniana* has a single columellar tooth in a markedly angular aperture.

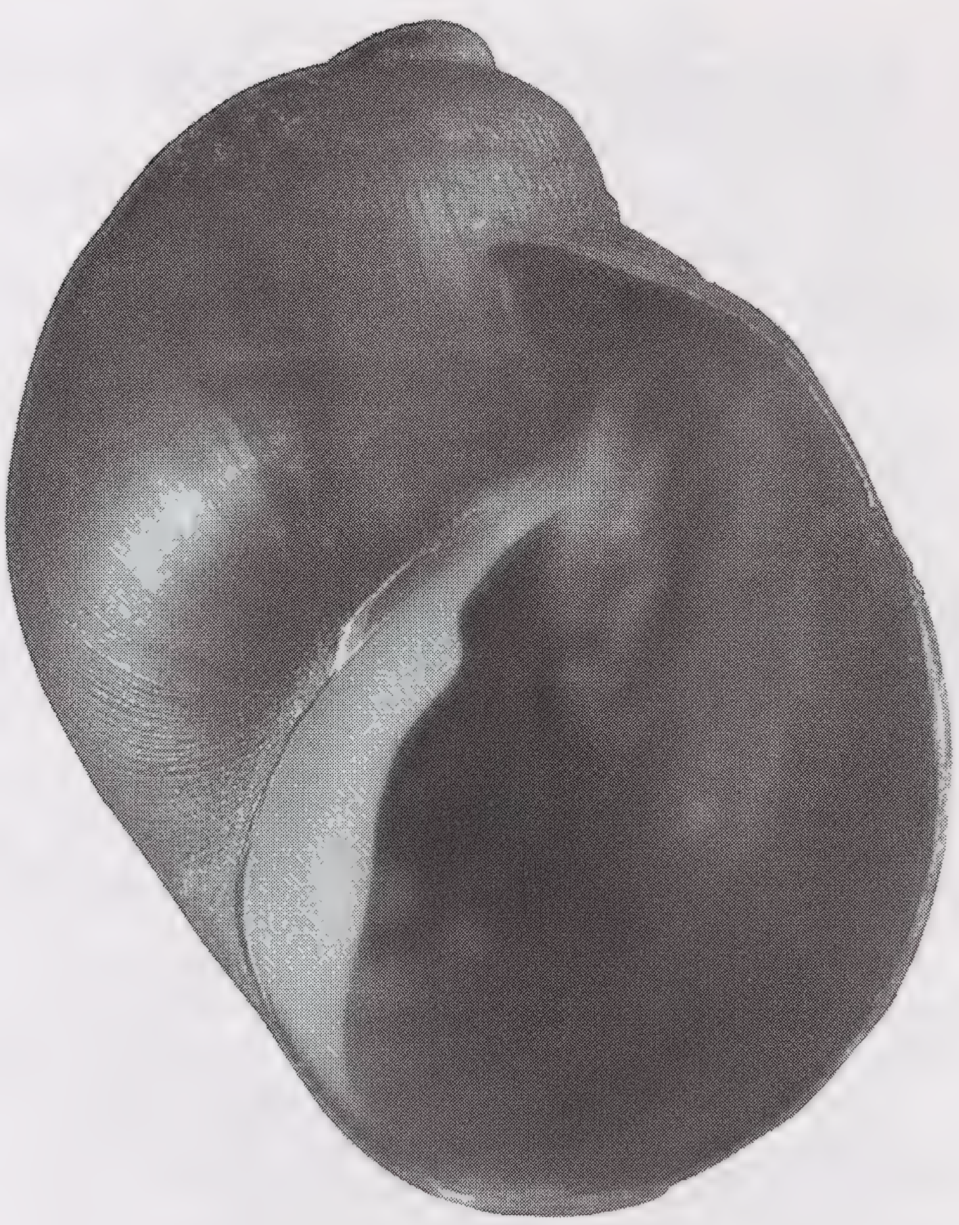

FIG. 8. Shell of *Chilina megastoma* from Arrechea Falls, Iguazú National Park (TL = 13.3 mm).

Another species with close geographic distribution, *Chilina parva* Martens, 1868, inhabits the states of Santa Catarina and Rio Grande do Sul, Brazil (Simone, 2006). This species is much smaller (5–8mm), with slight spire development and only one columellar tooth (Simone, 2006).

Chilina megastoma was compared with the new species because its internal anatomy is known, and its geographical distribution is closest to that of ***C. iguazuensis***. With respect to shell characters, aperture length is not equal to length of last whorl in *C. megastoma*, whereas ***C. iguazuensis*** has the same length; *C. megastoma* has two teeth on the labium and greater spire development, and it lacks interior iridescence, whereas ***C. iguazuensis*** has just one teeth in its labium, the spire is immersed, and it is iridescent within. It is similar to the new species in the possession of greatly developed aperture and reticulate shell. Maximum recorded size is 24.12 mm for ***C. iguazuensis***, 17.7 mm for *C. megastoma*.

Regarding anatomical traits, the two species differ in radular characteristics. ***Chilina iguazuensis*** has 58 tooth rows, with 51 teeth per half-row; the central tooth has serrated cusps, and the second lateral tooth is tetracuspid; *C. megastoma* has approximately 40 tooth rows with 42 teeth per half-row, the central tooth lacks serrated cusps, and the second lateral tooth is tricuspid.

Concerning the female reproductive system, the secondary bursa copulatrix of ***C. iguazuensis*** is uniformly wide along its entire length, whereas it expands terminally in *C. megastoma*. The course of the bursa copulatrix duct is similar in both species. Concerning the male reproductive system, prepuce length represents 60% of the length of the penis sheath in ***C. iguazuensis***, whereas it is 30 to 50% of penis sheath length in *C. megastoma*.

The nervous system shows the pattern described by Ituarte (1997) for *C. megastoma* regarding the number of connectives, but with differences in length. The connectives of *C. megastoma* are generally longer, relative to length of last whorl, than in ***C. iguazuensis***, except for the left pleuroparietal connective (Table 3). According to Ituarte (1997), in *C. megastoma* the length of the left cerebropleural connective is similar to that of the pleuroparietal connectives. However, our own dissections of relaxed specimens of this species indicate that the length of the left cerebropleural connective (11.29 ratio) is slightly more than twice that of the left pleuroparietal connectives (5.27 ratio). The left pleuroparietal and cerebropleural connectives are similar in ***C. iguazuensis***. According to Ituarte (1997), the left parietal-subintestinal connective of *C. megastoma* is approximately three times as long as the pleuroparietal connective, but our own dissections indicate a 4.4 ratio for these structures. This proportion is lower in ***C. iguazuensis*** (2.8 ratio). In addition, a nerve arising at approximately two-thirds of the length of the left parietal-subintestinal connective, which was mentioned by Ituarte (1997) and observed in our dissections of *C. megastoma*, was not detected in ***C. iguazuensis***.

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